REFLEX CARDIAC AND RESPIRATORY INHIBITION IN THE ELASMOBRANCH, *SCYLLIUM CANICULA*

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In the higher vertebrates the effect of stimulation of sensory nerves on the medullary centers is rather complex since either acceleration or inhibition of the heart may be obtained, often associated with acceleration or inhibition of respiration. In mammals Brodie and Russell (1900) found that stimulation of the pulmonary afferent fibers is more effective in causing reflex inhibition of the heart than excitation of afferent cardiac fibers, and the visceral supply is least effective. Bainbridge (1920), working also on mammals, obtained no reflex cardio-inhibition from the afferent pulmonary fibers, but did get marked respiratory inhibition. He found no evidence that the cardio-inhibitory center and the respiratory center are associated except through blood pressure changes. Alterations in blood pressure in the medulla have sometimes obscured true reflex processes. Thus Anrep and Starling (1925) offered evidence to show that inhibition of the heart in mammals could not be obtained reflexly through afferent fibers from the heart and aorta, and pointed out that an increase in blood pressure in the medulla caused cardiac slowing. These results contradicted in part the conclusions of Eyster and Hooker (1908), who believed that not only increased blood pressure in the medulla caused cardio-inhibition, but that reflex slowing of the heart could arise from the aorta, with both afferent and efferent pathways in the vagus. In 1926, however, Anrep and Segall confirmed the latter authors. Heymans (1929) has found cardio-inhibition in dogs to arise reflexly from increased blood pressure in the carotid sinus.

A survey of the literature on fishes, both cartilaginous and bony, gives one the impression that there is a nervous association between the respiratory rate and the heart rate. Schoenlein and Willem (1894) said that cessation of respiration in *Scyllium canicula* and *Torpedo ocellata* caused a reflex from the pharyngeal cavity which inhibited the heart. Bethe (1903) and Baglioni (1907) have found respiratory and cardiac inhibition occurring together in selachians. Lyon, (1926) studying blood pressure and respiration in sand sharks (*Carcharias*), noted that a great variety of stimuli applied externally or to certain viscera caused inhibition of the heart and some sort of respiratory response. He pointed out an intimate relationship between the respiratory rate and the heart rate through the vagus.

Since neither vasomotor nerves nor accelerator fibers have been demonstrated in elasmobranchs (Schoenlein, 1895; Bottazzi, 1902; Müller and Liljestrand, 1918; Lutz, 1930*a*), one might expect to find in these fishes an association between cardiac and respiratory inhibition without the complicating blood pressure factors found in mammals. In elasmobranchs it is also possible, because of the persistence of the functions of the medullary centers without a blood supply, to investigate the influence of stimulation of sensory fibers from the heart without involving blood pressure changes.

Material and Method

Vigorous specimens of the dogfish, *Scyllium canicula,* averaging 300 grams, taken during the months of March and April from the Bay of Naples, were used. One specimen of *Scyllium catulus* (625 grams) was put through the usual procedures carried out on *S. canicula* and was found to respond in a similar fashion, but the smaller species was more easily handled and more abundant. The fish was removed from the water and the forebrain was separated from the rest of the brain by a transverse cut just in front of the optic lobes and destroyed. The spinal cord was transected at a desired level and pithed posteriorly. Transection at a high level, namely, between the fifth and sixth vertebrae or even more anterior, was found desirable, when the details of the experiment permitted, in order to keep the fish quiet. It was then placed ventral side up on a fish holder and the gills were perfused through the mouth. Respiratory movements soon became regular. The heart was exposed through the ventral wall of the pericardial chamber and a small gold hook was put through the superficial tissue at the tip of the ventricle. By means of a thread and a light writing lever its movements were recorded. A similar hook passed through the anterior border of the third or fourth gill slit was attached to a thread, passed horizontally to a pulley, and up to a light writing lever for recording respiration.

When necessary the vagus and its branches, including the beginning of the lateral line nerve and the last four branchial nerves, and the hypobranchial nerve were exposed quickly by cutting a slit with the scissors through the skin beginning above the fifth gill slit about 3 mm. ventral to the lateral line and running anteriorly toward a point just above the posterior angle of the eye. This opened into the fascia between the dorsal musculature and the gills and directly over the anterior cardinal sinus, which was slit lengthwise exposing the nerves on its floor. This involved some loss of blood, and care was taken to keep the fish in a horizontal position to prevent sucking air into the heart. A bloodless method, described by Hemmeter (1912), of exposing "the ramus cardiacus" was found not to be practicable on the 300 gram fish, especially since the heart of *Scyllium canicula* receives not only a branch from the visceral trunk of the vagus but also a branch from the fifth branchial nerve (4th branchial division of the vagus).

The first large sympathetic ganglion was exposed when necessary by opening the abdominal cavity. Since this ganglion lies within the posterior cardinal sinus, the latter must be opened with considerable unavoidable loss of blood.

Results

Scyllium canicula was found to be very resistant to experimental procedures out of water when the gills were perfused. A preparation with the brain destroyed anterior to the optic lobes, the cord pithed posterior to the third vertebra, and the pericardial chamber opened, gave good cardiac and respiratory reflexes for over twenty-four hours. The laboratory temperature was about 14° C. When the anterior cardinal sinuses were opened, involving considerable bleeding, the

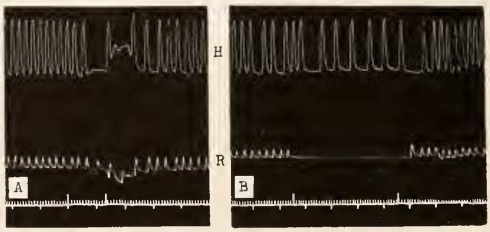

Fig. 1. Reflex cardiac and respiratory inhibition after transection of the ventral aorta. Forebrain and cord from the fifth vertebra destroyed. In this and subsequent figures the uppermost tracing is the heart record (*H*), the middle is respiration (*R*), and the lowest is the signal and time record. The large divisions are ten seconds. *A*, right fin pinched 21 minutes after transection of the aorta. *B*, gill perfusion stopped and started 31 minutes after transection.

preparation remained experimentally useful for about two hours. If in addition to the above procedures the abdominal cavity was entered and the posterior cardinal sinuses opened, medullary reflexes could be obtained for over an hour. In the latter case bleeding was profuse. When the aorta was transected, the respiratory center and the cardio-inhibitory center remained functional for over one hour (Fig. 1). Constant perfusion of the gills was found necessary to insure a quiet preparation, but without perfusion and with an intact circulation the

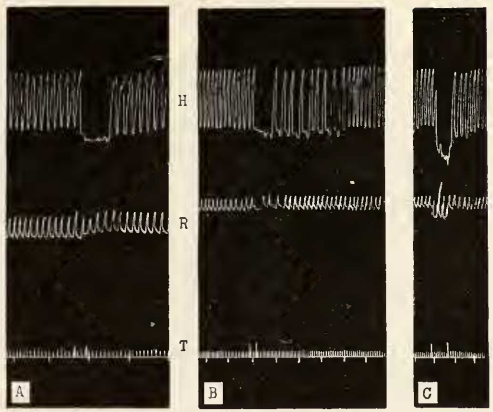

Fig. 2

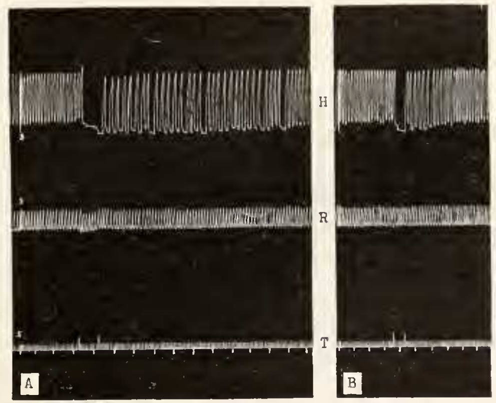

Fig. 3

Fig. 2. Cord pithed from the fifth vertebra. *A*, snout pinched gently with the fingers. Note reflex cardiac inhibition without respiratory inhibition. *B*, snout pinched vigorously. *C*, second right gill slit spread with forceps.

Fig. 3. Reflex cardiac inhibition without respiratory response. Cord pithed from the fifth vertebra. *A*, sides pinched with forceps dorsal to the pelvic fins. *B*, skin pinched on mid-abdomen.

medullary centers showed inhibitory reflexes for over three and one-half hours.

Cardiac and respiratory arrest or slowing were obtained with the cord pithed posteriorly from the second vertebra on mechanical and

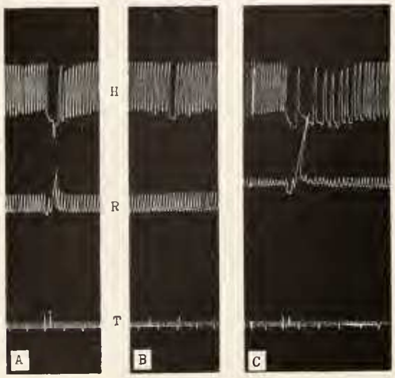

FIG. 4. Cord pithed from the fifth vertebra. *A,* needle stimulus to the left wall of the pericardial chamber. *B,* needle stimulus to the posterior wall. *C,* faradic stimulation of the posterior wall.

faradic stimulation of the skin of the head, nasal organs, gills, pectoral fins, cut surface of the coraco-mandibular muscle, pericardial walls, surface of the ventricle, oesophagus, stomach, spiral valve, and mesentery (Figs. 2, 3, 4 and 5). With the entire cord intact, similar

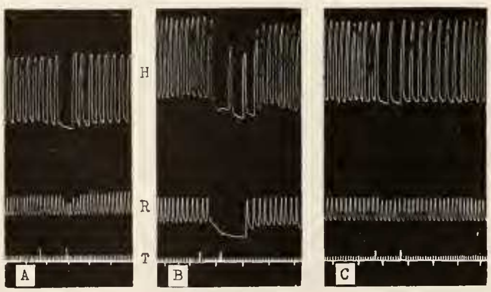

FIG. 5 Cord pithed from the third vertebra. *A,* stomach handled. *B,* posterior end of the stomach pinched with forceps. *C,* spiral valve pinched with forceps.

responses could be obtained from the kidney, epididymis, ovary, and uterus. No response could be elicited from the liver or testis even with the entire cord intact.

Faradic stimulation of the central end of the cut vagus, either the cardiac branch of the visceral ramus, the visceral ramus itself, or the cardiac branch of the fourth branchial division, the central end of the cut hypobranchial nerve, and the lateral line nerve (Fig. 6) produced cardiac and respiratory inhibition.

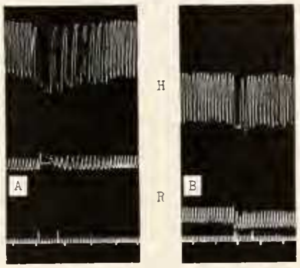

Fig. 6. Cord pithed from the eleventh vertebra. *A,* faradic stimulation of the central end of the left lateral line nerve. *B,* stimulus reduced.

Reflex cardiac inhibition and a reflex respiratory response were obtained when the rate of flow of the water to the gills was suddenly altered. The immediate response to starting and stopping the flow or to increasing and decreasing suddenly the rate was respiratory inhibi-

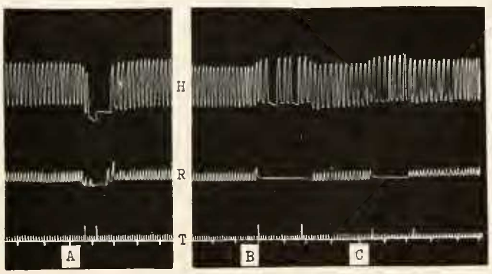

Fig. 7. Cord pithed from the fifth vertebra. *A,* water to the gills turned suddenly off and on. *B,* rate of flow of water increased and decreased to original rate. *C,* water gently stopped and started. Note respiratory inhibition without cardiac response.

tion, usually followed by one or more rapid and vigorous contractions of the gill muscles. The heart generally, but not always, showed inhibition as well (Fig. 7). This response occurred also after the aorta had been cut to prevent any possible blood pressure effects through changes in water pressure on the gill vessels.

When the sensory stimulation was strong or prolonged, both cardiac and respiratory inhibition occurred. Each effect, however, could be produced independently. If the stimulus was weak or brief, especially when applied to the skin, stomach, or spiral valve, only a cardiac response was elicited (Figs. 2*A*, 3, 4*B*, 5*C* and 6*B*). But a weak stimulus to the gill region frequently produced only respiratory inhibition. This occurred sometimes when the flow of perfusion water was altered (Fig. 7*C*).

Mechanical stimulation of the sides of the pericardial chamber by stroking with a sharp needle gave marked cardiac inhibition (Fig. 4). Weak faradic stimulation of the posterior wall of the chamber for a few seconds frequently gave an inhibitory effect lasting a half minute or more. On the other hand, pinching the sides of the body at the level of the pectoral fins vigorously with the forceps for twenty-five seconds stopped the heart for a short period, but it escaped from complete inhibition while still being reflexly stimulated.

When the surface of the ventricle was pinched lightly with small forceps, it immediately contracted and then became completely inhibited in diastole for 30 seconds accompanied by inhibition of respiration in relaxation and by movements of the head. If the ventricle was allowed to beat against a sharp needle, it contracted quickly a few times almost stopping in systole, then became completely inhibited in diastole, generally associated with respiratory inhibition. If the needle was inserted carefully under the epicardium, there was no systolic inhibition or respiratory response, but complete diastolic inhibition and slow recovery. Reflex diastolic inhibition from mechanical stimulation of the ventricle was also obtained after the aorta had been cut. A weak brief faradic stimulus applied to the surface of the ventricle caused immediate fibrillation under the electrode and, when the stimulus was removed, marked diastolic inhibition. Reflex cardio-inhibition from the ventricle was also obtained with only the post-branchial branches of the fourth branchial divisions intact. With the vagi cut or the heart excised, mechanical or faradic stimulation of either the auricle or the ventricle did not inhibit in diastole, but faradic stimulation along the Cuverian duct was immediately effective in producing this type of inhibition.

Discussion

While the results reported above indicate that respiratory and cardiac inhibition usually occur together, they show no evidence that there is a dependence of the one on the other. Each may occur independently. Moderate or strong stimulation, however, at any point mentioned above, except the liver and the testis, invariably caused both cardiac and respiratory inhibition. The liver and testis apparently have no sensory supply, stimulation of which gives either cardiac or respiratory responses.

Although stimulation of the central end of the lateral line nerve cut either at the gill region or midway between the pectoral and pelvic fins, gave both inhibitory responses, neither MacWilliam (1885) nor Kolff (1908) obtained heart reflexes on stimulation of this nerve in teleosts. Parker (1909) noted a temporary cessation of respiration on applying pressure to the lateral line region of the elasmobranch *Mustelus canis.*

The cardio-inhibition obtained on mechanical or electrical stimulation of the surface of the ventricle was undoubtedly of reflex nature since it was often accompanied by respiratory inhibition and skeletal muscular activity, responses which in themselves indicate afferent fibers from the ventricle. None of these responses was obtained with all of the cardiac branches of the vagi cut. In a paper on the innervation of the heart of *Scyllium* (Lutz, 1930*a*), further evidence is presented which indicates that the afferent pathway is in the vagus.

That reflex cardiac and respiratory inhibition should be so easily evoked by sensory stimulation of almost every part of *Scyllium* is interesting in view of the well-developed chromaphil system found in elasmobranchs and the inhibitory effect of adrenaline on the perfused heart of *Scyllium* described by Macdonald (1925). A similar effect of adrenalin chloride on a sinus-auricle preparation of various species of skate (*Raia*) and the dogfish, *Squalus acanthias,* has been described by Lutz (1930*b*). In view of the poorly developed sympathetic system (Müller and Liljestrand, 1918) and the lack of vaso-constrictor nerves (Schoenlein and Willem, 1894, and Schoenlein, 1895) found in elasmobranchs, the writer suggests that these inhibitory responses may be the expression of an emergency function which serves the fish in case of injury, especially to the gill region, and he has discussed the idea elsewhere (Lutz, 1930*b*).

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Summary

1. Mechanical and electrical stimulation of various parts of the surface of the body and certain viscera resulted in cardiac and respiratory inhibition when the spinal cord was pithed posterior to the second vertebra in *Scyllium canicula.*

2. Faradic stimulation of the central end of the cut vagus, either

the cardiac branches or the visceral ramus, and the central ends of the cut hypobranchial and lateral line nerves produced reflex cardiac and respiratory inhibition.

3. Stopping or starting the perfusion water through the gills or suddenly altering its rate of flow resulted in respiratory inhibition and sometimes cardiac inhibition.

4. Mechanical or electrical stimulation of the surface of the ventricle produced reflex cardio-inhibition and sometimes respiratory inhibition.

5. When stimulation was strong or prolonged, both cardiac and respiratory inhibition occurred, but weak stimulation of the gills or pharynx frequently produced only respiratory inhibition, whereas weak stimulation of the skin or viscera often produced only cardiac inhibition.

6. Both cardiac and respiratory inhibition were obtained for over twenty-four hours when the gills were perfused; for over three and one-half hours without perfusion; and for over one hour with the aorta transected at the heart. These phenomena, therefore, are true reflexes and are not due to alterations in blood pressure in the medulla.

7. The emergency function of the inhibitory responses is pointed out.

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